# Homework #3

*Problem #1 – The Power Stroke Model*

The Power Stroke Model is a mechanical model of actomyosin interaction developed by Sir Andrew Huxley in order to quantitatively understand actin-myosin force generation. Earlier you explored the polymerization of actin. The polymerization of actin is helical and the pitch of this helix has been measured as Δ = 36 nm. This means the polymer completes one turn of the helix every 36 nm along its length. Each of these actin subunits has only one binding site for myosin heads. The distance a myosin molecule stretches when it is cocked in preparation for a power stroke, δ, is only ~ 5 nm. Thus, the myosin molecule can only occur once in each full twist. However, the idea here is that many of these molecules work together to produce a sliding. Thus, myosin molecules link adjacent actin polymers together (cross-bridge model). We also know this process is regulated by ATP/ADP.

We can estimate the duty ratio or the time myosin is engaged, as

$$r_{duty} = \delta/\Delta = 5 \text{ nm} / 36 \text{ nm} = 0.14$$

However, it is know that myosin actually doesn't even bind this frequently and this is based on the force generation capacity of the entire actin-myosin bundle. So then, let's consider the force generation by stating the following:

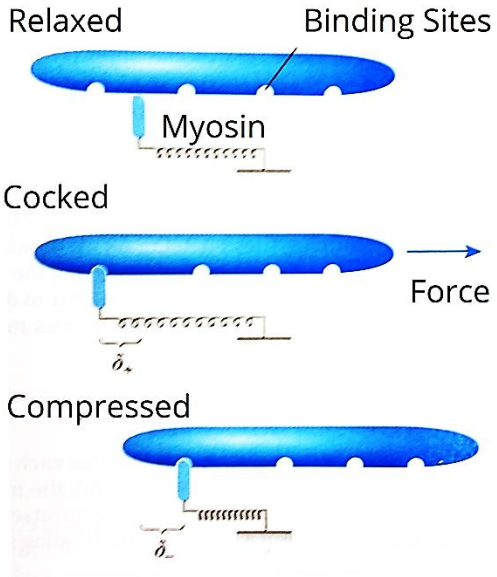
$$< F > = \frac{t_{on} < F_{on} > + t_{off} < F_{off} >}{t_{on} + t_{off}}$$

Here, we are saying the time average of the force is dependent on the on and off rates. However, the force generated when myosin is not engaged to actin must be zero $<F_{off}> = 0$. Thus, this simplifies:

$$< F > = \frac{t_{on}}{t_{on} + t_{off}} < F_{on} > = r_{duty} < F_{on} >$$

$$r_{duty} = \frac{\delta}{\Delta} = \frac{t_{on}}{t_{on} + t_{off}} = \frac{< F >}{< F_{on} >}$$

Now we have an idea of engagement of myosin with actin. Let's consider the power stroke model itself. We will treat the myosin heads like linear springs with some spring constant $k_m$. The model considers three possible positions (as seen below).




The spring moves forward by a distance $\delta_+$ when it is cocked and imparts a force of $k_m\delta_+$ on the actin filament. In the "compressed position," the spring compresses by $\delta_-$ past the zero position before releasing acting. **Based on this illustration, can you figure out the average force $<F_{on}>$?**

With the expression you wrote down for force, the limiting condition below, and the previous expressions derived, **rewrite the expression for <F> in terms of $k_m$, $\delta_+$, $\delta_-$, and $\Delta$.**

$$\delta = \delta_+ + \delta_-$$

You should have derived a positive and negative term, or $F_+$ and $F_-$.
Let's see if we can use what you derived to relate it to sliding velocity. What does the positive term actually mean? It depends on distance that the myosin head move forward when cocked. At this point, most myosin heads are bound so there is no reason to expect that this would depend on velocity. The second term depends on distance the heads overshoot the zero force position before releasing. Thus, it seems reasonable to assume this compression distance depends on sliding velocity. Let's introduce a new parameter for release time, $t_r$. **Rewrite the <F> expression for sliding velocity. You can keep the first term as $F_+$.**

**Now, let's examine the force-velocity behavior. Which features are consistent with the Hill model? Which are not?**

Let's resolve the issue. Consider Δ, the distance between myosin binding sites on actin. Let's call the minimum distance $d_s$. From the duty ratio argument, we expect Δ to exceed $d_s$ by some amount but the myosin head and actin-binding site must align eventually. Therefore,

$$\Delta = nd_s \; where \; n \; is \; an \; integer.$$

To estimate what n might be, consider some effective sweet spot of width $w_{bs}$. The time available for binding within this width is inversely related to velocity.

$$t = \frac{w_{bs}}{v}$$

Further, we assume that while the myosin is in this sweet spot, it will bind actin at a constant on rate of $k_+$. **Write the rate equation for [M] based on previous lectures.**

**Solve this differential equation, which was done in class to understand the Bell model for adhesive bonds.**

What you just solved is from the perspective of an individual myosin molecule and you can use it to define the probability of transitioning from an unbound to bound state in the time available for binding. **Do this here.**

**From this expression, what happens to the probability when the velocity is too high?**

Consider another limiting case, Δ>>d and n is large. [Relatively few of the binding sites are occupied]. Because each binding site is separated from the next by a distance δ and the distance between an occupied site is Δ, **then the probability of a given head being bound is also given by…**

**Set your two expressions for probability equal to each other and solve for Δ.**

Replace Δ in your expression for <F>. **Does this resemble Hill's model?** Try out the following parameters.
$k_m$ = 5 pN/nm.
t_ = 0.6 ms.
$d_s$ = 36 nm
$\delta_+$ = 5 nm.
$k_+$ = 21 $s^{-1}$ [this really depends on ATP concentration but this is theoretical max).
$w_{bs}$ = 36 nm [this is hard to measure but theoretical max is the $d_s$]
**For these parameters, plot the curve.** You'll find it is not precisely hyperbolic but within physiological range.

*Problem #2 – Coupled Neurons Simulation*

In class, we derived the equation for the FitzHugh-Nagumo Model. The FitzHugh-Nagumo model is a simplified version of the Hodgkin-Huxley model. Recall, it was a nonlinear system. I am going to provide Matlab code online that explores the dynamics of two FitzHugh neurons coupled via linear coupling. The equations are the following:

$$\frac{dv_1}{dt} = -{v_1}^3 + (1 + a_1){v_1}^2 - a_1 v_1 - r_1 + I + d_{12} v_2$$

$$\frac{dr_1}{dt} = b v_1 - c r_1$$

$$\frac{dv_2}{dt} = -{v_2}^3 + (1 + a_2){v_2}^2 - a_2 v_2 - r_2 + I + d_{21} v_1$$

$$\frac{dr_2}{dt} = b v_2 - c r_2$$

There are two pieces to the code. One is the function for these equations and the second is for you to plot and change parameters. Initial conditions set are [$v_1(0)$, $v_2(0)$] = [0.01, 0.01] and [ $r_1(0)$, $r_2(0)$] = [0, 0]. Parameters set are $a_1$ = 0.05, $a_2$ = 0.25, b=c= 0.01 and I = 0.1.

Set the interaction parameters to ($d_{12}$, $d_{21}$): (0,0), (0,0.2), (-0.1,0.2), (-0.3,0.2), (-.5,0.2), and compare and contrast the time series plots and the phase plot. What conclusions can you draw based on these simulations? As a bit of bonus, feel free to expand on this problem and explore all the other parameters and their effect.

*Problem # 3 - Choose either the Engler paper or Spatz paper and answer the questions regarding to each paper.*

For the 2006 manuscript "Matrix elasticity directs stem cell lineage specification"…

1. Summarize the manuscript in no more than 150 words.
2. What are the three cell types discussed in this manuscript? Make a table to compare (i) their elastic stiffnesses, (ii) their microstructural appearances, and (iii) their cellular functions. Feel free to consult other sources of information to complement the table, i.e., cell images from the web, etc.
3. Discuss the impact of the major findings in this manuscript on stem cell therapies. As a typical example, you might think of the direct injection of undifferentiated human embryonic stem cells into the infarcted region of the heart. In response to the infarct, compliant contracting heart muscle cells are replaced by stiff scar tissue. What are the dangers of stem cell injection therapies in view of the experimental results of the manuscript?

For the 2014 manuscript "Nanoparticle Tension Probes Patterned at the Nanoscale: Impact Integrin Clustering on Force Transmission" …

1. Summarize the manuscript in no more than 150 words.
2. What is a focal adhesion and its role in different cellular processes? What conclusion is reached about the formation of focal adhesions and force generation?
3. If you could use this technology, what would you study and why?

*Problem #4 – Mechanotransduction Gone Awry*

The recent review "Mechanotransduction gone awry" by Jaalouk and Lammerding discusses defects in mechanotransduction and their effects on various different disease types.

1. Read the manuscript carefully and summarize it in approximately 150 words.
2. Select your favorite mechanotransduction pathway and describe how it is altered under diseased conditions.
3. What are the implications of this article? Write a paragraph of about 150 words.